



Epstein–Barr Virus and Rheumatoid Arthritis: Is There a Link?

Citation

Costenbader, Karen H., and Elizabeth W. Karlson. 2006. Epstein–Barr virus and rheumatoid arthritis: Is there a link? *Arthritis Research & Therapy* 8(1): 204.

Published Version

doi://10.1186/ar1893

Permanent link

<http://nrs.harvard.edu/urn-3:HUL.InstRepos:10021571>

Terms of Use

This article was downloaded from Harvard University's DASH repository, and is made available under the terms and conditions applicable to Other Posted Material, as set forth at <http://nrs.harvard.edu/urn-3:HUL.InstRepos:dash.current.terms-of-use#LAA>

Share Your Story

The Harvard community has made this article openly available.
Please share how this access benefits you. [Submit a story](#).

[Accessibility](#)

Review

Epstein-Barr virus and rheumatoid arthritis: is there a link?

Karen H Costenbader and Elizabeth W Karlson

Brigham and Women's Hospital, Division of Rheumatology, Immunology and Allergy, Department of Medicine, Harvard Medical School, 75 Francis Street, Boston, MA 02115, USA

Corresponding author: Karen H Costenbader, kcostenbader@partners.org

Published: 16 January 2006

This article is online at <http://arthritis-research.com/content/8/1/204>

© 2006 BioMed Central Ltd

Arthritis Research & Therapy 2006, **8**:204 (doi:10.1186/ar1893)**Abstract**

Rheumatoid arthritis is a systemic autoimmune disease characterized by chronic, destructive, debilitating arthritis. Its etiology is unknown; it is presumed that environmental factors trigger development in the genetically predisposed. Epstein-Barr virus, a nearly ubiquitous virus in the human population, has generated great interest as a potential trigger. This virus stimulates polyclonal lymphocyte expansion and persists within B lymphocytes for the host's life, inhibited from reactivating by the immune response. In latent and replicating forms, it has immunomodulating actions that could play a role in the development of this autoimmune disease. The evidence linking Epstein-Barr virus and rheumatoid arthritis is reviewed.

Introduction

Rheumatoid arthritis (RA) is a chronic inflammatory polyarthritis that progressively destroys synovial joints and can cause systemic complications. RA affects about 1% of the world's population [1], and its prevalence in women is twofold to fourfold that in men [2,3]. RA has enormous personal, social, and economic impact [4,5]; women with RA have overall mortality rates 2.3-fold those in age-matched controls [6]. New biologic therapies, based on an increasing understanding of the molecular mechanisms involved in RA, afford a more normal life to many, but the burden of disease remains high. At present there is no known cure. Despite improved therapy, the long-term prognosis remains poor and average life expectancy is reduced by 3 to 18 years [7]. Both the direct costs of treatment of RA and the indirect costs of disability and loss from the workplace are high [8,9].

RA is marked by extensive synovial hyperplasia and infiltration by lymphocytes, monocytes, macrophages, and fibroblasts. RA is a predominantly CD4⁺ T helper type 1 (Th1)-driven disease [10]. Aberrant T cell activation is one of the earliest events in the development of RA, with CD4⁺ T cells stimulating monocytes and macrophages to produce

inflammatory cytokines, including interleukin (IL)-1, IL-6, and tumor necrosis factor- α (TNF- α), as well as proteolytic enzymes, destroying synovium, cartilage, and underlying bone [11]. The T cells infiltrating the rheumatoid synovium are oligoclonal, implicating an antigen-driven process [12,13], but the inciting antigen or antigens remain unidentified. Activated T cells also signal B cells to produce increased levels of immunoglobulins, including rheumatoid factor (RF). Autoreactive B cells also have a central role in the development of RA, producing autoantibodies that might be involved in tissue damage in RA [14].

Genetic factors are important in disease susceptibility, but environmental exposures are probably crucial as well. Many exposures have been investigated as possible risk factors for the development of RA, including reproductive factors such as the use of oral contraceptives, hormone replacement therapy, and breast feeding [15-17], and dietary factors such as antioxidants [18,19], red meat protein [20,21], and fat intake [22,23]. However, most of these have shown only weak associations. Cigarette smoking is the only exposure that has repeatedly been found to increase the risk of RA, with a relative risk of about 1.8 [24-27].

Viruses and the development of RA

A viral trigger of RA in the genetically predisposed has been hypothesized for many years [28-36]. A virus could act as an adjuvant in the development of autoimmunity, non-specifically stimulating innate immune responses, including mast cells, dendritic cells, Toll-like receptors and complement receptors [37]. Polyarthritis resembling RA is seen clinically soon after exposure to multiple viruses including rubella, human T cell leukemia virus-1 (HTLV-1), parvovirus B19, and hepatitis B and C [36,38-40]. Exposure to a common virus would explain the ubiquity of RA worldwide. However, such a virus has

CCP = cyclic citrullinated peptide; EA-D = Epstein-Barr virus early antigen – diffuse; EA-R = Epstein-Barr virus early antigen – restricted; EBNA = Epstein-Barr virus nuclear antigen; EBV = Epstein-Barr virus; HLA = human leukocyte antigen; HTLV-1 = human T cell leukemia virus-1; IL = interleukin; RA = rheumatoid arthritis; RF = rheumatoid factor; SLE = systemic lupus erythematosus; Th1 = T helper type 1; TNF = tumor necrosis factor; VCA = Epstein-Barr virus viral capsid antigen.

eluded identification by modern techniques possibly because of a long latency period, with RA onset years after initial exposure. Viruses including Epstein-Barr virus (EBV), parvovirus B19, HTLV-1, human herpesvirus-6, human herpesvirus-8, and human endogenous retroviruses-5 have all been proposed to be involved in the pathogenesis of RA [31-35,41-44].

However, most of the evidence implicating viruses in the pathogenesis of RA is circumstantial and inconclusive. Tantalizing observations have often been based on *in vitro* or animal studies, case reports, or studies with small sample sizes, cross-sectional designs, or no control groups.

Epstein-Barr virus

EBV, the causative agent of infectious mononucleosis, is a DNA-containing herpesvirus that is extremely prevalent worldwide, infecting more than 98% of the human population by the age of 40 years [45]. It is highly associated with several malignancies including nasopharyngeal carcinoma, Burkitt's lymphoma, T/NK cell lymphomas, lymphoproliferative disease in immunocompromised hosts, and Hodgkin's disease, in most of which EBV genomes are detectable within tumor cells [45].

To initiate infection, EBV uses its major envelope glycoprotein, gp350, to bind to its receptor, complement receptor-2, on epithelial cells and B lymphocytes [46,47]. Major histocompatibility complex (MHC) class II molecules are cofactors for the infection of B cells by EBV [48]. During initial infection there is massive polyclonal expansion of B lymphocytes, followed by that of CD8⁺ T lymphocytes in particular [49]. EBV then becomes latent within memory B lymphocytes and persists for the lifetime of its human host. While it is latent within the B cell, its viral genome is intact as an episome, but most viral genes are not active [50]. The proteins it produces are responsible for inhibiting apoptosis and blocking the antiviral effects of interferon- γ on EBV-transformed B cells [50,51]. EBV has multiple immunomodulating actions. Binding of its major envelope glycoprotein gp350 to complement receptor-2 leads to the upregulation of the important inflammatory cytokines IL-1 β , TNF- α , and IL-6 [52-54]. EBV encodes an immunosuppressive viral IL-10 cytokine and a viral colony-stimulating factor-1 cytokine receptor, involved in its ability to escape immune detection [55-57]. B cell transformation by EBV also induces the expression of EBV-induced gene 3 (*EBI3*), which encodes a form of IL-12, responsible for the initiation of Th1-type immunity [58-60].

The host's cellular immune response has primary responsibility for the control of latent EBV infection within the B cells [49,50]. CD4⁺ T cells activate the innate immune response to EBV and are required for the generation of robust memory responses by CD8⁺ cells, which is important in suppressing EBV [61-63]. EBV reactivation and EBV-related lymphoproliferative diseases occur in immunosuppressed renal and bone marrow transplant patients [64] and in association with HIV [65].

During late incubation and the early infectious phase of mononucleosis, antibodies against EBV viral capsid antigen (VCA) and early antigen complex – diffuse (EA-D) appear [45,66]. Later, weeks to months after disease onset, antibodies against EBV nuclear antigen (EBNA) and early antigen complex – restricted (EA-R) emerge (Table 1) [45,67]. Antibodies against EBNA-2 are detected first and decline within a few weeks, followed by the rise of EBNA-1 antibodies, which normally persist at a stable level for life [45,67]. Thus, in a normal adult, latent EBV infection is associated with moderate, stable and highly correlated levels of IgG antibodies against VCA, EBNA-1, and EA-R, with very low or undetectable levels of antibodies against EBNA-2 and EA-D [67-69].

In situations of decreased cellular immunity, however, EBV reactivation, or the transition from latent to lytic infection, can occur. Anti-VCA IgG antibodies, anti-EBNA-2 antibodies, and anti-EA antibodies are often elevated in these situations, which is consistent with EBV reactivation. The relationship between EBV serologic responses and levels of viral replication, as detected by polymerase chain reaction, is variable [45,69]. Latent EBV can replicate and spread despite the presence of antibodies, and antibody titers correlate with viral activity rather than with the degree of protection afforded [68]. In many diseases strongly associated with EBV, such as nasopharyngeal carcinoma and Burkitt's lymphoma, anti-EBV serologies are abnormal many years before the onset of disease. In nasopharyngeal carcinoma, for example, levels of IgA anti-VCA antibody 10-fold those in normal subjects are found years in advance of the onset of disease [70], indicative of high levels of viral replication. IgA anti-VCA antibody titers are used for screening in Asia, where nasopharyngeal cancer is endemic [71,72].

EBV and the pathogenesis of RA

In the quest to uncover an infectious trigger of RA, much research has concentrated on the potential for molecular mimicry presented by EBV. EBV was first implicated in the pathogenesis of RA by Alsbaugh and Tan [30,73], who reported that sera from patients with RA were reactive against a nuclear antigen in EBV-transformed lymphocytes. This 'RA nuclear antigen' was determined as a glycine/alanine-rich repeat in EBNA-1 [74,75]. Antibodies against this repeat are cross-reactive with a 62 kDa protein present in the synovium of patients with RA, but not in that of controls [76-78]. Antigenic sequence similarities exist between other EBV proteins and RA-specific proteins as well. These include the EBV-encoded protein gp110, which has sequence homology with the QKRAA amino acid motif (the 'shared epitope') of the β -chain of human leukocyte antigen (HLA)-DR4 [79,80]. Humans with EBV infection have antibodies against the gp110 protein, as well as T cells with receptors that recognize the QKRAA motif in both gp110 and HLA-DR4 molecules. In addition, antibodies against EBV peptide

Table 1**Patterns of anti-Epstein-Barr virus (EBV) serologies observed in different disease states**

Disease state	VCA	EBNA-1	EBNA-2	EA	References
Early, acute primary EBV infection	IgA, IgM	Undetectable	↑↑	EA-D	[45,66]
Primary infection (weeks to months)	IgG	↑	↓	EA-R	[45,67]
Latent EBV infection in healthy host	Stable IgG	Stable	↓	Stable EA-R	[67]
Reactivation/EBV replication	↑↑ IgG	↑↑	↑	↑	[67-69,117]
Nasopharyngeal carcinoma ^a	↑↑ IgG, IgA			↑ IgG, IgA	[70,118-120]
Burkitt's lymphoma ^a	↑↑ IgG	↑	↑↑	↑	[70,121]
Multiple sclerosis ^a	↑	↑	↑↑	↑	[111,112]
Systemic lupus erythematosus ^a	↑	↑			[113,114]
Rheumatoid arthritis	↑	↑	↑	↑	[82-86,122]

EA, EBV early antigen; EA-D, EBV early antigen – diffuse; EA-R, EBV early antigen – restricted; EBNA, EBV nuclear antigen; VCA, EBV viral capsid antigen. ^aAbnormalities observed before disease onset.

p107, the major epitope of the EBV-encoded EBNA-1 antigen, recognize and bind to denatured collagen and keratin [81]. These findings support the hypothesis that molecular mimicry, either by influencing T cell receptor recognition of the HLA 'shared epitope' or through the production of autoantibodies against joint proteins, is involved in RA disease pathogenesis.

Patients with existing RA have higher levels of antibodies against several EBV-encoded proteins, including VCA [82], early antigen (EA) [82], EBNA-1 [82-85], and EBNA-2 [86], than do healthy controls, and the presence of RF does not seem to be related to these elevations (Table 1). Patients suffering from RA have a 10-fold increase in EBV DNA load in peripheral blood mononuclear cells compared with that in controls; this elevation is stable and not influenced by the presence or absence of RF, age, duration of RA, disease activity, or RA treatment [87]. Patients with RA have significantly higher numbers of circulating EBV-infected B cells [88] and EBV DNA loads in saliva [42]. Several studies have shown that levels of EBV DNA and mRNA are much higher in the synovium of patients with RA than in that of healthy controls [83,89-91]. Synovial EBV DNA loads are highest in patients with RA with at least one copy of the HLA-DRB1 'shared epitope', the strongest known genetic risk factor for RA [89]. However, these cross-sectional findings have never been tested in a prospective cohort with blood drawn before the diagnosis of RA. Nevertheless, given the ubiquity of the virus in the population, a binary assay for the presence of anti-EBV antibodies years preceding the onset of RA would be less informative than a sensitive titer quantification compared with controls.

EBV-specific T cell function is also impaired in RA [92-98]. A large proportion of the CD8⁺ T cells infiltrating rheumatoid synovium recognize the EBV transactivating factors, BZLF-1

and BMLF-1, important in the control of EBV reactivation [99]. The HLA-DR4 shared epitope, a strong genetic risk factor for RA, is associated with low frequencies of T cells specific for the EBV gp110 glycoprotein, also critical in the control of EBV infection [98]. Clonal expansion of peripheral CD8⁺ CD28⁻ EBV-specific T cells is observed in patients with RA but not in controls [100]. These cells are thought to be dysfunctional, senescent suppressor T cells, possibly caused by recurrent EBV stimulation and/or a primary defect of T cell differentiation and proliferation in RA.

Antibodies directed against cyclic citrullinated peptides (CCPs) are increasingly important in the early diagnosis of RA [101,102]. Citrullination is the process of deimination of peptidyl arginine to peptidyl citrulline, recognized specifically by anti-CCP antibodies. These autoantibodies are directed against citrullinated proteins in the rheumatoid synovium, including fibrin, filaggrin, perinuclear factor, and keratin [103]. They are highly specific for RA (sensitivity 68%, specificity 98%) [101] and in prospective cohort studies are present several years before the onset of RA [104-106]. Klareskog and colleagues in Sweden have found that cigarette smoking may trigger HLA-DR restricted immune reactions to autoantigens modified by citrullination, potentially explaining the interaction between HLA shared epitope and cigarette smoking that greatly increases the risk of anti-CCP-positive RA (L Klareskog, personal communication). Although it has not yet been studied in relation to the citrullination of autoantigens or the formation of autoantibodies, EBV could potentially have a similar role. Moreover, the regulation of B cell apoptosis might be important in the production of anti-CCP antibodies [107]; EBV persists indefinitely in host B cells and encodes at least two proteins that interfere with apoptosis, namely BHFR1 (a viral homologue of the anti-apoptotic protein Bcl-2) [108] and LMP-1 (latent membrane protein-1) [109].

Chicken or egg?

Although the observations noted above support an association between EBV, or the host's immune response to it, and RA, this association need not be causative. Elevated anti-EBV antibody titers have also been found in other autoimmune diseases, including Sjögren's syndrome [110], and years before the onset of both multiple sclerosis [111,112] and systemic lupus erythematosus (SLE) [113,114]. Anti-EBV antibody titers rise gradually from their first detectable levels years before the first symptoms of SLE until the time of SLE diagnosis, paralleling, and in some cases preceding, the development of SLE-specific antibodies [113,114].

Whether the observed abnormalities in EBV-directed immune responses and EBV viral loads are a cause or a consequence of RA remains a mystery. Through its potential for molecular mimicry, by polyclonal activation of B cells, or via some other mechanism, EBV or an EBV-specific immune response could be a trigger for the development of RA in the genetically predisposed. Alternatively, an innate or acquired immune defect in those with or at risk for RA could handicap the host's ability to suppress this chronic viral infection. There is mounting evidence that patients with lupus, for example, have impaired EBV-specific immune responses [115] and the frequency of EBV-infected cells in the blood of patients with SLE increases during SLE disease flares, independently of immunosuppressive therapy and in concert with aberrant expression of viral proteins [116]. This suggests that in those with SLE, and perhaps similarly in those with RA, T cell control of latent EBV infection is defective. Whether the virus actually has an etiologic role in these autoimmune diseases, or whether underlying immune abnormalities allow dysregulation of latent EBV as an epiphenomenon, is the crux of the matter.

Conclusion

The cause of RA, a highly disabling systemic autoimmune disease, remains unknown. Family studies and genome-wide scans have shown that there is an important genetic influence in the susceptibility to RA; evidence points to a common virus, such as EBV, that could act as a trigger in genetically susceptible hosts. So far, studies looking for an association between EBV infection and RA have been characterized by small numbers and retrospective or cross-sectional designs. Patients with established RA seem to have elevated levels of anti-EBV antibodies and viral loads. These study designs have not been able to address the timing of these abnormalities with regard to the development of RA, nor have they been able to exclude the possibility that RA itself, or its treatment, is responsible for abnormally elevated EBV serologic responses and viral loads. Understanding the timing and directionality of the EBV-RA relationship is crucial to distinguishing inciting from secondary events in RA pathogenesis and to advancing our understanding of the etiology of RA.

Competing interests

The author(s) declare that they have no competing interests.

Acknowledgements

The authors acknowledge the contributions of Dr Frederick C-S Wang in writing this review. KHC is the recipient of an American College of Rheumatology/Arthritis Foundation Arthritis Investigator Award.

References

- Gabriel SE, Crowson CS, O'Fallon WM: **The epidemiology of rheumatoid arthritis in Rochester, Minnesota, 1955-1985.** *Arthritis Rheum* 1999, **42**:415-420.
- Lincoff A, Worthington JW, O'Fallon WM, Kurland LT: **The epidemiology of rheumatoid arthritis in Rochester, Minnesota: a study of incidence, prevalence, and mortality.** *Am J Epidemiol* 1980, **111**:87-98.
- Symmons DP, Barrett EM, Bankhead CR, Scott DG, Silman AJ: **The incidence of rheumatoid arthritis in the United Kingdom: results from the Norfolk Arthritis Register.** *Br J Rheumatol* 1994, **33**:735-739.
- Yelin E, Henke C, Epstein W: **The work dynamics of the person with rheumatoid arthritis.** *Arthritis Rheum* 1987, **30**:507-512.
- Yelin E: **The costs of rheumatoid arthritis: absolute, incremental, and marginal estimates.** *J Rheumatol Suppl* 1996, **44**:47-51.
- Wolfe F, Mitchell DM, Sibley JT, Fries JF, Bloch DA, Williams CA, Spitz PW, Hays RD, Kleinheksel SM, Cathey MA: **The mortality of rheumatoid arthritis.** *Arthritis Rheum* 1994, **37**:481-494.
- Pincus T, Callahan LF: **Taking mortality in rheumatoid arthritis seriously—predictive markers, socioeconomic status and comorbidity.** *J Rheumatol* 1986, **13**:841-845.
- Yelin E, Wanke LA: **An assessment of the annual and long-term direct costs of rheumatoid arthritis: the impact of poor function and functional decline.** *Arthritis Rheum* 1999, **42**:1209-1218.
- McIntosh E: **The cost of rheumatoid arthritis.** *Br J Rheumatol* 1996, **35**:781-790.
- Miossec P, van den Berg W: **Th1/Th2 cytokine balance in arthritis.** *Arthritis Rheum* 1997, **40**:2105-2115.
- Choy EH, Panayi GS: **Cytokine pathways and joint inflammation in rheumatoid arthritis.** *N Engl J Med* 2001, **344**:907-916.
- Goronzy JJ, Bartz-Bazzanella P, Hu W, Jendro MC, Walser-Kuntz DR, Weyand CM: **Dominant clonotypes in the repertoire of peripheral CD4+ T cells in rheumatoid arthritis.** *J Clin Invest* 1994, **94**:2068-2076.
- Waase I, Kayser C, Carlson PJ, Goronzy JJ, Weyand CM: **Oligoclonal T cell proliferation in patients with rheumatoid arthritis and their unaffected siblings.** *Arthritis Rheum* 1996, **39**:904-913.
- Kotzin BL: **The role of B cells in the pathogenesis of rheumatoid arthritis.** *J Rheumatol Suppl* 2005, **73**:14-18.
- Hernandez-Avila M, Liang MH, Willett WC, Stampfer MJ, Colditz GA, Rosner B, Chang RW, Hennekens CH, Speizer FE: **Exogenous sex hormones and the risk of rheumatoid arthritis.** *Arthritis Rheum* 1990, **33**:947-953.
- Hernandez-Avila M, Liang MH, Willett WC, Stampfer MJ, Colditz GA, Rosner B, Roberts WN, Hennekens CH, Speizer FE: **Reproductive factors, smoking, and the risk for rheumatoid arthritis.** *Epidemiology* 1990, **1**:285-291.
- Karlson EW, Mandl LA, Hankinson SE, Grodstein F: **Do breastfeeding and other reproductive factors influence future risk of rheumatoid arthritis? Results from the Nurses' Health Study.** *Arthritis Rheum* 2004, **50**:3458-3467.
- Jaswal S, Mehta HC, Sood AK, Kaur J: **Antioxidant status in rheumatoid arthritis and role of antioxidant therapy.** *Clin Chim Acta* 2003, **338**:123-129.
- Kamanli A, Naziroglu M, Aydilek N, Hacıevliyagil C: **Plasma lipid peroxidation and antioxidant levels in patients with rheumatoid arthritis.** *Cell Biochem Funct* 2004, **22**:53-57.
- Pattison DJ, Symmons D, Luben R, Welch A, Khaw KT, Day NJ, Silman AJ: **High red meat and total protein consumption are risk factors for new onset inflammatory polyarthritis: results from a population-based prospective study [abstract].** *Arthritis Rheum* 2003, **48**:S393.
- Grant WB: **The role of meat in the expression of rheumatoid arthritis.** *Br J Nutr* 2000, **84**:589-595.

22. Shapiro JA, Koepsell TD, Voigt LF, Dugowson CE, Kestin M, Nelson JL: **Diet and rheumatoid arthritis in women: a possible protective effect of fish consumption.** *Epidemiology* 1996, **7**: 256-263.
23. Linos A, Kaklamani VG, Kaklamani E, Koumantaki Y, Giziaki E, Papazoglou S, Mantzoros CS: **Dietary factors in relation to rheumatoid arthritis: a role for olive oil and cooked vegetables?** *Am J Clin Nutr* 1999, **70**:1077-1082.
24. Karlson EW, Lee IM, Cook NR, Manson JE, Buring JE, Hennekens CH: **A retrospective cohort study of cigarette smoking and risk of rheumatoid arthritis in female health professionals.** *Arthritis Rheum* 1999, **42**:910-917.
25. Hutchinson D, Shepstone L, Moots R, Lear JT, Lynch MP: **Heavy cigarette smoking is strongly associated with rheumatoid arthritis (RA), particularly in patients without a family history of RA.** *Ann Rheum Dis* 2001, **60**:223-227.
26. Matthey DL, Dawes PT, Fisher J, Brownfield A, Thomson W, Hajeer AH, Ollier WE: **Nodular disease in rheumatoid arthritis: association with cigarette smoking and HLA-DRB1/TNF gene interaction.** *J Rheumatol* 2002, **29**:2313-2318.
27. Stolt P, Bengtsson C, Nordmark B, Lindblad S, Lundberg I, Klareskog L, Alfredsson L: **Quantification of the influence of cigarette smoking on rheumatoid arthritis: results from a population based case-control study, using incident cases.** *Ann Rheum Dis* 2003, **62**:835-841.
28. Albert LJ, Inman RD: **Molecular mimicry and autoimmunity.** *N Engl J Med* 1999, **341**:2068-2074.
29. Bonneville M, Scotet E, Peyrat MA, Saulquin X, Houssaint E: **Epstein-Barr virus and rheumatoid arthritis.** *Rev Rhum Engl Ed* 1998, **65**:365-368.
30. Alsbaugh MA, Jensen FC, Rabin H, Tan EM: **Lymphocytes transformed by Epstein-Barr virus. Induction of nuclear antigen reactive with antibody in rheumatoid arthritis.** *J Exp Med* 1978, **147**:1018-1027.
31. Cooke SP, Rigby SP, Griffiths DJ, Venables PJ: **Viral studies in rheumatic disease.** *Ann Med Interne (Paris)* 1998, **149**:30-33.
32. Dostal C, Newkirk MM, Duffy KN, Paleckova A, Bosak V, Cerna M, Zdar'sky E, Zvarova J: **Herpes viruses in multicase families with rheumatoid arthritis and systemic lupus erythematosus.** *Ann N Y Acad Sci* 1997, **815**:334-337.
33. Inman RD: **Infectious etiology of rheumatoid arthritis.** *Rheum Dis Clin North Am* 1991, **17**:859-870.
34. Carty SM, Snowden N, Silman AJ: **Should infection still be considered as the most likely triggering factor for rheumatoid arthritis?** *J Rheumatol* 2003, **30**:425-429.
35. Griffiths DJ: **Rheumatoid arthritis: a viral aetiology?** *Hosp Med* 2000, **61**:378-379.
36. Masuko-Hongo K, Kato T, Nishioka K: **Virus-associated arthritis.** *Best Pract Res Clin Rheumatol* 2003, **17**:309-318.
37. Fairweather D, Frisanchi-Kiss S, Rose NR: **Viruses as adjuvants for autoimmunity: evidence from Coxsackievirus-induced myocarditis.** *Rev Med Virol* 2005, **15**:17-27.
38. Phillips PE: **Viral arthritis.** *Curr Opin Rheumatol* 1997, **9**:337-344.
39. Ytterberg SR: **Viral arthritis.** *Curr Opin Rheumatol* 1999, **11**:275-280.
40. Nades SJ: **Rheumatic manifestations of parvovirus B19 infection.** *Rheum Dis Clin North Am* 1998, **24**:375-401.
41. Kerr JR: **Pathogenesis of human parvovirus B19 in rheumatic disease.** *Ann Rheum Dis* 2000, **59**:672-683.
42. Newkirk MM, Watanabe Duffy KN, Leclerc J, Lambert N, Shiroky JB: **Detection of cytomegalovirus, Epstein-Barr virus and herpes virus-6 in patients with rheumatoid arthritis with or without Sjogren's syndrome.** *Br J Rheumatol* 1994, **33**:317-322.
43. Meyer O: **Parvovirus B19 and autoimmune diseases.** *Joint Bone Spine* 2003, **70**:6-11.
44. Zhang L, Nikkari S, Skurnik M, Ziegler T, Luukkainen R, Mottonen T, Toivanen P: **Detection of herpesviruses by polymerase chain reaction in lymphocytes from patients with rheumatoid arthritis.** *Arthritis Rheum* 1993, **36**:1080-1086.
45. Linde A: **Epstein-Barr virus.** In *Manual of Clinical Microbiology*. Vol. 2. 8th edition. Edited by Murray PR, Baron EJ, Jorgensen JH, Pfaller MA, Tenover FC. Washington DC: ASM Press; 2003:1331-1340.
46. Fingerroth JD, Weis JJ, Tedder TF, Strominger JL, Biro PA, Fearon DT: **Epstein-Barr virus receptor of human B lymphocytes is the C3d receptor CR2.** *Proc Natl Acad Sci USA* 1984, **81**:4510-4514.
47. Nemerow GR, Mold C, Schwend VK, Tollefson V, Cooper NR: **Identification of gp350 as the viral glycoprotein mediating attachment of Epstein-Barr virus (EBV) to the EBV/C3d receptor of B cells: sequence homology of gp350 and C3 complement fragment C3d.** *J Virol* 1987, **61**:1416-1420.
48. Li Q, Spriggs MK, Kovats S, Turk SM, Comeau MR, Nepom B, Hutt-Fletcher LM: **Epstein-Barr virus uses HLA class II as a cofactor for infection of B lymphocytes.** *J Virol* 1997, **71**:4657-4662.
49. Rickinson AB, Moss DJ: **Human cytotoxic T lymphocyte responses to Epstein-Barr virus infection.** *Annu Rev Immunol* 1997, **15**:405-431.
50. Cohen JL: **Epstein-Barr virus infection.** *N Engl J Med* 2000, **343**: 481-492.
51. Kieff E: **Epstein-Barr virus and its replication.** In *Fields Virology*. Vol. 2. Edited by Fields BN, Knipe DM, Howley PM. Philadelphia: Lippincott, Williams & Wilkins; 2001:2511-2575.
52. Krauer KG, Belzer DK, Liaskou D, Buck M, Cross S, Honjo T, Sculley T: **Regulation of interleukin-1 β transcription by Epstein-Barr virus involves a number of latent proteins via their interaction with RBP.** *Virology* 1998, **252**:418-430.
53. D'Addario M, Ahmad A, Xu JW, Menezes J: **Epstein-Barr virus envelope glycoprotein gp350 induces NF- κ B activation and IL-1 β synthesis in human monocytes-macrophages involving PKC and PI3-K.** *FASEB J* 1999, **13**:2203-2213.
54. D'Addario M, Ahmad A, Morgan A, Menezes J: **Binding of the Epstein-Barr virus major envelope glycoprotein gp350 results in the upregulation of the TNF-alpha gene expression in monocytic cells via NF- κ B involving PKC, PI3-K and tyrosine kinases.** *J Mol Biol* 2000, **298**:765-778.
55. Moore KW, Vieira P, Fiorentino DF, Trounstein ML, Khan TA, Mosmann TR: **Homology of cytokine synthesis inhibitory factor (IL-10) to the Epstein-Barr virus gene BCRF1.** *Science* 1990, **248**:1230-1234.
56. Hsu DH, de Waal Malefyt R, Fiorentino DF, Dang MN, Vieira P, de Vries J, Spits H, Mosmann TR, Moore KW: **Expression of interleukin-10 activity by Epstein-Barr virus protein BCRF1.** *Science* 1990, **250**:830-832.
57. Cohen JL, Lekstrom K: **Epstein-Barr virus BRF1 protein is dispensable for B-cell transformation and inhibits alpha interferon secretion from mononuclear cells.** *J Virol* 1999, **73**: 7627-7632.
58. Yoshimoto T, Nagase H, Yoneto T, Inoue J, Nariuchi H: **Interleukin-12 expression in B cells by transformation with Epstein-Barr virus.** *Biochem Biophys Res Commun* 1998, **252**: 556-560.
59. Devergne O, Birkenbach M, Kieff E: **Epstein-Barr virus-induced gene 3 and the p35 subunit of interleukin 12 form a novel heterodimeric hematopoietin.** *Proc Natl Acad Sci USA* 1997, **94**: 12041-12046.
60. Holscher C: **The power of combinatorial immunology: IL-12 and IL-12-related dimeric cytokines in infectious diseases.** *Med Microbiol Immunol (Berl)* 2004, **193**:1-17.
61. Williams H, McAulay K, Macsween KF, Gallacher NJ, Higgins CD, Harrison N, Swerdlow AJ, Crawford DH: **The immune response to primary EBV infection: a role for natural killer cells.** *Br J Haematol* 2005, **129**:266-274.
62. Schoenberger SP, Toes RE, van der Voort EI, Offringa R, Melief CJ: **T-cell help for cytotoxic T lymphocytes is mediated by CD40-CD40L interactions.** *Nature* 1998, **393**:480-483.
63. Bennett SR, Carbone FR, Karamalis F, Flavell RA, Miller JF, Heath WR: **Help for cytotoxic-T-cell responses is mediated by CD40 signalling.** *Nature* 1998, **393**:478-480.
64. Kaden J, Petersen S, Kaden K, May G: **Epstein-Barr virus infection after kidney transplantation.** *Transpl Int* 1998, **11** Suppl 1: S119-S124.
65. Ragona G, Sirianni MC, Soddu S, Vercelli B, Sebastiani G, Piccoli M, Aiuti F: **Evidence for dysregulation in the control of Epstein-Barr virus latency in patients with AIDS-related complex.** *Clin Exp Immunol* 1986, **66**:17-24.
66. Linde A, Andersson J, Lundgren G, Wahren B: **Subclass reactivity to Epstein-Barr virus capsid antigen in primary and reactivated EBV infections.** *J Med Virol* 1987, **21**:109-121.
67. Henle W, Henle G, Andersson J, Ernberg I, Klein G, Horwitz CA, Marklund G, Rymo L, Wellinder C, Straus SE: **Antibody**

- responses to Epstein-Barr virus-determined nuclear antigen (EBNA)-1 and EBNA-2 in acute and chronic Epstein-Barr virus infection. *Proc Natl Acad Sci USA* 1987, **84**:570-574.
68. Linde A: **Diagnosis of Epstein-Barr virus-related diseases.** *Scand J Infect Dis Suppl* 1996, **100**:83-88.
69. Lennette ET, Rymo L, Yadav M, Masucci G, Merk K, Timar L, Klein G: **Disease-related differences in antibody patterns against EBV-encoded nuclear antigens EBNA 1, EBNA 2 and EBNA 6.** *Eur J Cancer* 1993, **29A**:1584-1589.
70. Seigneurin JM, Lavoue MF, Genoulaz O, Bornkamm GW, Lenoir GM: **Antibody response against the Epstein-Barr virus-coded nuclear antigen2 (EBNA2) in different groups of individuals.** *Int J Cancer* 1987, **40**:349-353.
71. Kantakamalakul W, Chongkolwatana C, Naksawat P, Muangsomboon S, Sukpanichnant S, Chongvisal S, Metheetrairat C, Kositanont U, Puthavathana P: **Specific IgA antibody to Epstein-Barr viral capsid antigen: a better marker for screening nasopharyngeal carcinoma than EBV-DNA detection by polymerase chain reaction.** *Asian Pac J Allergy Immunol* 2000, **18**:221-226.
72. Ng WT, Yau TK, Yung RW, Sze WM, Tsang AH, Law AL, Lee AW: **Screening for family members of patients with nasopharyngeal carcinoma.** *Int J Cancer* 2005, **113**:998-1001.
73. Alspaugh MA, Tan EM: **Serum antibody in rheumatoid arthritis reactive with a cell-associated antigen. Demonstration by precipitation and immunofluorescence.** *Arthritis Rheum* 1976, **19**: 711-719.
74. Baboonian C, Halliday D, Venables PJ, Pawlowski T, Millman G, Maini RN: **Antibodies in rheumatoid arthritis react specifically with the glycine alanine repeat sequence of Epstein-Barr nuclear antigen-1.** *Rheumatol Int* 1989, **9**:161-166.
75. Petersen J, Rhodes G, Roudier J, Vaughan JH: **Altered immune response to glycine-rich sequences of Epstein-Barr nuclear antigen-1 in patients with rheumatoid arthritis and systemic lupus erythematosus.** *Arthritis Rheum* 1990, **33**:993-1000.
76. Rumpold H, Rhodes GH, Bloch PL, Carson DA, Vaughan JH: **The glycine-alanine repeating region is the major epitope of the Epstein-Barr nuclear antigen-1 (EBNA-1).** *J Immunol* 1987, **138**:593-599.
77. Kouri T, Petersen J, Rhodes G, Aho K, Palosuo T, Heliövaara M, Isomaki H, von Essen R, Vaughan JH: **Antibodies to synthetic peptides from Epstein-Barr nuclear antigen-1 in sera of patients with early rheumatoid arthritis and in preillness sera.** *J Rheumatol* 1990, **17**:1442-1449.
78. Fox R, Sportsman R, Rhodes G, Luka J, Pearson G, Vaughan J: **Rheumatoid arthritis synovial membrane contains a 62,000-molecular-weight protein that shares an antigenic epitope with the Epstein-Barr virus-encoded associated nuclear antigen.** *J Clin Invest* 1986, **77**:1539-1547.
79. Roudier J, Petersen J, Rhodes GH, Luka J, Carson DA: **Susceptibility to rheumatoid arthritis maps to a T-cell epitope shared by the HLA-Dw4 DR β -1 chain and the Epstein-Barr virus glycoprotein gp110.** *Proc Natl Acad Sci USA* 1989, **86**:5104-5108.
80. Lotz M, Roudier J: **Epstein-Barr virus and rheumatoid arthritis: cellular and molecular aspects.** *Rheumatol Int* 1989, **9**:147-152.
81. Birkenfeld P, Haratz N, Klein G, Sulitzeanu D: **Cross-reactivity between EBNA 1p107 peptide, collagen and keratin: implications for the pathogenesis of rheumatoid arthritis.** *Clin Immunol Immunopathol* 1990, **54**:14-25.
82. Ferrell PB, Aitchison CT, Pearson GR, Tan EM: **Seroepidemiological study of relationships between Epstein-Barr virus and rheumatoid arthritis.** *J Clin Invest* 1981, **67**:681-687.
83. Blaschke S, Schwarz G, Moneke D, Binder L, Muller G, Reuss-Borst M: **Epstein-Barr virus infection in peripheral blood mononuclear cells, synovial fluid cells, and synovial membranes of patients with rheumatoid arthritis.** *J Rheumatol* 2000, **27**:866-873.
84. Catalano MA, Carson DA, Slovin SF, Richman DD, Vaughan JH: **Antibodies to Epstein-Barr virus-determined antigens in normal subjects and in patients with seropositive rheumatoid arthritis.** *Proc Natl Acad Sci USA* 1979, **76**:5825-5828.
85. Alspaugh MA, Henle G, Lennette ET, Henle W: **Elevated levels of antibodies to Epstein-Barr virus antigens in sera and synovial fluids of patients with rheumatoid arthritis.** *J Clin Invest* 1981, **67**:1134-1140.
86. Hazelton RA, Sculley TB, Pope JH: **The prevalence of antibodies to an Epstein-Barr virus-induced polypeptide (EBNA-2) in the sera of rheumatoid arthritic families.** *Br J Rheumatol* 1987, **26**:193-196.
87. Balandraud N, Meynard JB, Auger I, Sovran H, Mugnier B, Revirion D, Roudier J, Roudier C: **Epstein-Barr virus load in the peripheral blood of patients with rheumatoid arthritis: accurate quantification using real-time polymerase chain reaction.** *Arthritis Rheum* 2003, **48**:1223-1228.
88. Tosato G, Steinberg AD, Yarchoan R, Heilman CA, Pike SE, De Seau V, Blaese RM: **Abnormally elevated frequency of Epstein-Barr virus-infected B cells in the blood of patients with rheumatoid arthritis.** *J Clin Invest* 1984, **73**:1789-1795.
89. Saal JG, Krimmel M, Steidle M, Gerneth F, Wagner S, Fritz P, Koch S, Zacher J, Sell S, Einsele H, Muller CA: **Synovial Epstein-Barr virus infection increases the risk of rheumatoid arthritis in individuals with the shared HLA-DR4 epitope.** *Arthritis Rheum* 1999, **42**:1485-1496.
90. Takei M, Mitamura K, Fujiwara S, Horie T, Ryu J, Osaka S, Yoshino S, Sawada S: **Detection of Epstein-Barr virus-encoded small RNA 1 and latent membrane protein 1 in synovial lining cells from rheumatoid arthritis patients.** *Int Immunol* 1997, **9**:739-743.
91. Takeda T, Mizugaki Y, Matsubara L, Imai S, Koike T, Takada K: **Lytic Epstein-Barr virus infection in the synovial tissue of patients with rheumatoid arthritis.** *Arthritis Rheum* 2000, **43**: 1218-1225.
92. Bardwick PA, Bluestein HG, Zvaifler NJ, Depper JM, Seegmiller JE: **Altered regulation of Epstein-Barr virus induced lymphoblast proliferation in rheumatoid arthritis lymphoid cells.** *Arthritis Rheum* 1980, **23**:626-632.
93. Depper JM, Bluestein HG, Zvaifler NJ: **Impaired regulation of Epstein-Barr virus-induced lymphocyte proliferation in rheumatoid arthritis is due to a T cell defect.** *J Immunol* 1981, **127**:1899-1902.
94. Tosato G, Steinberg AD, Blaese RM: **Defective EBV-specific suppressor T-cell function in rheumatoid arthritis.** *N Engl J Med* 1981, **305**:1238-1243.
95. Gaston JS, Rickinson AB, Epstein MA: **Epstein-Barr virus-specific cytotoxic T cell responses in rheumatoid arthritis patients.** *Rheumatol Int* 1982, **2**:155-159.
96. Stierle HE, Brown KA, Pery JD, Holborow EJ: **Increased responsiveness of rheumatoid B lymphocytes to stimulation by Epstein-Barr virus.** *Rheumatol Int* 1983, **3**:7-11.
97. Kahan A, Amor B, Menkes CJ: **Different defects of T cell regulation of Epstein-Barr virus-induced B cell activation in rheumatoid arthritis.** *Arthritis Rheum* 1985, **28**:961-970.
98. Toussiot E, Wendling D, Tiberghien P, Luka J, Roudier J: **Decreased T cell precursor frequencies to Epstein-Barr virus glycoprotein Gp110 in peripheral blood correlate with disease activity and severity in patients with rheumatoid arthritis.** *Ann Rheum Dis* 2000, **59**:533-538.
99. Scotet E, David-Ameline J, Peyrat MA, Moreau-Aubry A, Pinczon D, Lim A, Even J, Semana G, Berthelot JM, Breathnach R, et al.: **T cell response to Epstein-Barr virus transactivators in chronic rheumatoid arthritis.** *J Exp Med* 1996, **184**:1791-1800.
100. Klatt T, Ouyang Q, Flad T, Koetter I, Buhning HJ, Kalbacher H, Pawelec G, Muller CA: **Expansion of peripheral CD8+ CD28- T cells in response to Epstein-Barr virus in patients with rheumatoid arthritis.** *J Rheumatol* 2005, **32**:239-251.
101. Schellekens GA, Visser H, de Jong BA, van den Hoogen FH, Hazes JM, Breedveld FC, van Venrooij WJ: **The diagnostic properties of rheumatoid arthritis antibodies recognizing a cyclic citrullinated peptide.** *Arthritis Rheum* 2000, **43**:155-163.
102. Bizzaro N, Mazzanti G, Tonutti E, Villalta D, Tozzoli R: **Diagnostic accuracy of the anti-citrulline antibody assay for rheumatoid arthritis.** *Clin Chem* 2001, **47**:1089-1093.
103. Baeten D, Peene I, Union A, Meheus L, Sebbag M, Serre G, Veys EM, De Keyser F: **Specific presence of intracellular citrullinated proteins in rheumatoid arthritis synovium: relevance to antiagggrin autoantibodies.** *Arthritis Rheum* 2001, **44**:2255-2262.
104. Rantapää-Dahlqvist S, de Jong BA, Berglin E, Hallmans G, Wadell G, Stenlund H, Sundin U, van Venrooij WJ: **Antibodies against cyclic citrullinated peptide and IgA rheumatoid factor predict the development of rheumatoid arthritis.** *Arthritis Rheum* 2003, **48**:2741-2749.

105. Berglin E, Padyukov L, Sundin U, Hallmans G, Stenlund H, Van Venrooij WJ, Klareskog L, Dahlqvist SR: **A combination of autoantibodies to cyclic citrullinated peptide (CCP) and HLA-DRB1 locus antigens is strongly associated with future onset of rheumatoid arthritis.** *Arthritis Res Ther* 2004, **6**:R303-R308.
106. Nielen MM, van Schaardenburg D, Reesink HW, van de Stadt RJ, van der Horst-Bruinsma IE, de Koning MH, Habibuw MR, Vandenbroucke JP, Dijkmans BA: **Specific autoantibodies precede the symptoms of rheumatoid arthritis: a study of serial measurements in blood donors.** *Arthritis Rheum* 2004, **50**:380-386.
107. Lopez-Hoyos M, Marquina R, Tamayo E, Gonzalez-Rojas J, Izui S, Merino R, Merino J: **Defects in the regulation of B cell apoptosis are required for the production of citrullinated peptide autoantibodies in mice.** *Arthritis Rheum* 2003, **48**:2353-2361.
108. Henderson S, Huen D, Rowe M, Dawson C, Johnson G, Rickinson A: **Epstein-Barr virus-coded BHRF1 protein, a viral homologue of Bcl-2, protects human B cells from programmed cell death.** *Proc Natl Acad Sci USA* 1993, **90**:8479-8483.
109. Kulwichit W, Edwards RH, Davenport EM, Baskar JF, Godfrey V, Raab-Traub N: **Expression of the Epstein-Barr virus latent membrane protein 1 induces B cell lymphoma in transgenic mice.** *Proc Natl Acad Sci USA* 1998, **95**:11963-11968.
110. Yamazaki M, Kitamura R, Kusano S, Eda H, Sato S, Okawa-Takatsui M, Aotsuka S, Yanagi K: **Elevated immunoglobulin G antibodies to the proline-rich amino-terminal region of Epstein-Barr virus nuclear antigen-2 in sera from patients with systemic connective tissue diseases and from a subgroup of Sjogren's syndrome patients with pulmonary involvements.** *Clin Exp Immunol* 2005, **139**:558-568.
111. Ascherio A, Munger KL, Lennette ET, Spiegelman D, Hernan MA, Olek MJ, Hankinson SE, Hunter DJ: **Epstein-Barr virus antibodies and risk of multiple sclerosis: a prospective study.** *JAMA* 2001, **286**:3083-3088.
112. Levin LI, Munger KL, Rubertone MV, Peck CA, Lennette ET, Spiegelman D, Ascherio A: **Temporal relationship between elevation of Epstein-Barr virus antibody titers and initial onset of neurological symptoms in multiple sclerosis.** *JAMA* 2005, **293**:2496-2500.
113. McClain MT, Bruner TL, Dennis GJ, Harley JB, James JA: **The temporal relationship between the onset of anti-EBNA-1 and lupus autoimmunity supports a role for EBV in the development of SLE [abstract].** *Arthritis Rheum* 2003, **48**:S674.
114. Heinlen LD, McClain MT, Dennis GJ, Rubertone MV, Harley JB, James JA: **The development of antibodies targeting Epstein-Barr virus closely parallels autoimmune progression near the onset of SLE [abstract].** *Arthritis Rheum* 2003, **48**:S662.
115. Kang I, Quan T, Nolasco H, Park SH, Hong MS, Crouch J, Pamer EG, Howe JG, Craft J: **Defective control of latent Epstein-Barr virus infection in systemic lupus erythematosus.** *J Immunol* 2004, **172**:1287-1294.
116. Gross AJ, Hochberg D, Rand WM, Thorley-Lawson DA: **EBV and systemic lupus erythematosus: a new perspective.** *J Immunol* 2005, **174**:6599-6607.
117. Obel N, Hoier-Madsen M, Kangro H: **Serological and clinical findings in patients with serological evidence of reactivated Epstein-Barr virus infection.** *Apmis* 1996, **104**:424-428.
118. Henle G, Henle W: **Epstein-Barr virus-specific IgA serum antibodies as an outstanding feature of nasopharyngeal carcinoma.** *Int J Cancer* 1976, **17**:1-7.
119. Neel HB, Pearson GR, Weiland LH, Taylor WF, Goepfert HH, Pilch BZ, Goodman M, Lanier AP, Huang AT, Hyams VJ, et al.: **Application of Epstein-Barr virus serology to the diagnosis and staging of North American patients with nasopharyngeal carcinoma.** *Otolaryngol Head Neck Surg* 1983, **91**:255-262.
120. Ringborg U, Henle W, Henle G, Ingimarsson S, Klein G, Silfversward C, Strander H: **Epstein-Barr virus-specific serodiagnostic tests in carcinomas of the head and neck.** *Cancer* 1983, **52**:1237-1243.
121. Nkrumah F, Henle W, Henle G, Herberman R, Perkins V, Depue R: **Burkitt's lymphoma: its clinical course in relation to immunologic reactivities to Epstein-Barr virus and tumor-related antigens.** *J Natl Cancer Inst* 1976, **57**:1051-1056.
122. Shimizu N, Yamaki M, Sakuma S, Ono Y, Takada K: **Three Epstein-Barr virus (EBV)-determined nuclear antigens induced by the BamHI E region of EBV DNA.** *Int J Cancer* 1988, **41**:744-751.